

APPENDIX B
PENDING CLAIMS AS OF OCTOBER 12, 2001
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27. A combinatorial gene expression library, comprising a pool of expression constructs, each expression construct containing one or more cDNA or genomic DNA fragments, wherein the cDNA or genomic DNA fragments in the pool of expression constructs are derived from an environmental sample comprising a plurality of species of donor organisms, and wherein the cDNA or genomic DNA fragments in each expression construct are operably-associated each with one or more regulatory regions that drives expression of genes encoded by the cDNA or genomic DNA fragments in an appropriate host organism.

28. A combinatorial chimeric pathway gene expression library, comprising a pool of expression constructs, each expression construct containing randomly concatenated cDNA or genomic DNA fragments derived from an environmental sample comprising one or more species of donor organisms, in which the concatenated cDNA or genomic DNA fragments are operably-associated with one or more regulatory regions that drive expression of genes encoded by the concatenated cDNA or genomic DNA fragments in an appropriate host organism.

29. A biased combinatorial gene expression library, comprising a pool of expression constructs, each expression construct containing cDNA or genomic DNA fragments preselected from a plurality of species of donor organisms derived from an environmental sample for a specific property, in which the cDNA or genomic DNA

fragments are operably-associated with one or more regulatory regions that drive expression of genes encoded by the cDNA or genomic DNA fragments in an appropriate host organism.

30. The gene expression library of claim 27, 28, or 29 wherein the environmental sample is a soil sample.

31. The gene expression library of claim 27, 28, or 29 wherein the environmental sample is selected from the group consisting of deposits near hot springs, deposits near thermal vents, freshwater filtrates, marine sediments, estuarine sediments, or seawater filtrates.

32. The gene expression library of claim 27, 28, or 29 wherein at least one of the cDNA or genomic DNA fragments comprises nucleotide sequences that encode for proteins or fragments thereof that are involved in secondary metabolism.

33. The gene expression library of claim 27, 28, or 29 wherein at least one of the cDNA or genomic DNA fragments comprises nucleotide sequences that encode for proteins or fragments thereof that are involved in antibiotic biosynthesis.

34. The gene expression library of claim 27, 28, or 29 wherein at least one of the cDNA or genomic DNA fragments comprises nucleotide sequences that encode for proteins or fragments thereof that are involved in polyketide biosynthesis.

36. The gene expression library of claim 27, 28, or 29 wherein the expression constructs are contained in host cells.

37. The gene expression library of claim 30 wherein the expression constructs are contained in host cells.

38. The gene expression library of claim 31 wherein the expression constructs are contained in host cells.

39. The gene expression library of claim 32 wherein the expression constructs are contained in host cells.

40. The gene expression library of claim 33 wherein the expression constructs are contained in host cells.

41. The gene expression library of claim 34 wherein the expression constructs are contained in host cells.

43. The gene expression library of claim 27, 28, or 29 wherein the expression constructs comprises a plasmid vector, a phage vector, a viral vector, a cosmid vector, or an artificial chromosome.

44. A biased combinatorial gene expression library, comprising a pool of expression constructs, each expression construct containing cDNA or genomic DNA fragments some of which are preselected from a plurality of species of donor organisms by hybridization of the cDNA or genomic DNA fragments to nucleic acid probes comprising nucleotide sequences that encode for proteins or fragments thereof that are involved in secondary metabolism, in which the cDNA or genomic DNA fragments are operably

associated with one or more regulatory regions that drive expression of genes encoded by the cDNA or genomic DNA fragments in an appropriate host organism.

45. The biased combinatorial gene expression library of claim 44 wherein some of the cDNA or genomic DNA fragments are preselected by hybridization of the cDNA or genomic fragments to nucleic acid probes comprising nucleotide sequences that encode for proteins or fragments thereof that are involved in polyketide biosynthesis.

46. The biased combinatorial gene expression library of claim 44 wherein some of the cDNA or genomic DNA fragments are preselected by hybridization of the cDNA or genomic DNA fragments to nucleic acid probes comprising nucleotide sequences that encode for proteins or fragments thereof that are involved in antibiotic biosynthesis.

48. The biased combinatorial gene expression library of claim 44 wherein some of the cDNA or genomic DNA fragments are preselected by hybridization of the cDNA or genomic DNA fragments to nucleic acid probes comprising nucleotide sequences that encode for proteins or fragments thereof that are involved in the biosynthesis of erythromycin, actinorhodin, thiostrepton, virginiamycin, valinomycin, or actinomycin.

49. The biased combinatorial gene expression library of claim 44, 46, or 48 wherein the expression constructs are contained in host cells.

50. The biased combinatorial gene expression library of claim 44, 46, or 48 wherein the expression constructs comprise a plasmid vector, a phage vector, a viral vector, a cosmid vector, or an artificial chromosome.

51. The gene expression library of claim 27, 28, or 29 wherein at least one of the cDNA or genomic DNA fragments comprises nucleotide sequences that encode for proteins or fragments thereof that are involved in peptide biosynthesis.

52. The gene expression library of claim 27, 28, or 29 wherein at least one of the cDNA or genomic DNA fragments comprises nucleotide sequences that encode for proteins or fragments thereof that are involved in glycoside biosynthesis.

53 The gene expression library of claim 27, 28, or 29 wherein at least one of the cDNA or genomic DNA fragments comprises nucleotide sequences that encode for proteins or fragments thereof that are involved in aminoglycoside biosynthesis.

54. The gene expression library of claim 51 wherein the expression constructs are contained in host cells.

55. The gene expression library of claim 52 wherein the expression constructs are contained in host cells.

56. The gene expression library of claim 53 wherein the expression constructs are contained in host cells.

57. The biased combinatorial gene expression library of claim 44 wherein some of the cDNA or genomic DNA fragments are preselected by hybridization of the cDNA or genomic fragments to nucleic acid probes comprising nucleotide sequences that encode for proteins or fragments thereof that are involved in peptide biosynthesis.

58. The biased combinatorial gene expression library of claim 44 wherein some of the cDNA or genomic DNA fragments are preselected by hybridization of the cDNA or genomic fragments to nucleic acid probes comprising nucleotide sequences that encode for proteins or fragments thereof that are involved in glycoside biosynthesis.

59. The biased combinatorial gene expression library of claim 44 wherein some of the cDNA or genomic DNA fragments are preselected by hybridization of the cDNA or genomic fragments to nucleic acid probes comprising nucleotide sequences that encode for proteins or fragments thereof that are involved in aminoglycoside biosynthesis.

61. The biased combinatorial gene expression library of claim 45 wherein the expression constructs are contained in host cells.

62. The biased combinatorial gene expression library of claim 57 wherein the expression constructs are contained in host cells.

63. The biased combinatorial gene expression library of claim 58 wherein the expression constructs are contained in host cells.

64. The biased combinatorial gene expression library of claim 59 wherein the expression constructs are contained in host cells.

66. The biased combinatorial gene expression library of claim 45 wherein the expression constructs comprise a plasmid vector, a phage vector, a viral vector, a cosmid vector, or an artificial chromosome.

67. The biased combinatorial gene expression library of claim 57 wherein the expression constructs comprise a plasmid vector, a phage vector, a viral vector, a cosmid vector, or an artificial chromosome.

68. The biased combinatorial gene expression library of claim 58 wherein the expression constructs comprise a plasmid vector, a phage vector, a viral vector, a cosmid vector, or an artificial chromosome.

69. The biased combinatorial gene expression library of claim 59 wherein the expression constructs comprise a plasmid vector, a phage vector, a viral vector, a cosmid vector, or an artificial chromosome.